Final Report - Grant No: NAG10-0209
Sweetpotato Stem Cuttings Database in Preparation for Flight

Introduction

Since 1985, Tuskegee University has been engaged in research that addresses NASA's mission to achieve long-duration human space exploration on the moon and Mars. The successful research of Dr. George Washington Carver, the beginning of our long history of working with crops for food and industrial uses, is well known. This history and contemporary technical expertise in food/industrial crops, and our interest in pursuing new, interdisciplinary approaches to education and research led us to submit a proposal to NASA in 1985 to pursue the hydroponic production of sweetpotato for food for astronaut-explorers on future long-term, distant space missions. As a carbohydrate and dual vegetable (edible leaves), the versatile sweetpotato has the potential to be a key crop as a main source of energy and other nutrients such as beta-carotene, calcium and potassium. Sweetpotato is a crop very suited to space, as was documented in a monograph published by our scientists in 1984.

Stem cuttings are commonly used in propagating the sweetpotato. Because the stem cuttings regenerate easily and quickly, a study of their early growth and development in microgravity (or on clinostats) can prove quite useful in understanding the morphological changes that might occur under such conditions for any crop that can be regenerated from cuttings. Sweetpotato offers distinct advantages for this type of space related research. First, it is easier and faster to regenerate from a stem cutting and is more true to type than from seed, and second, its genetic makeup can be perpetuated from one initial planting. A database defining sweetpotato stem cutting growth in microgravity is consistent with NASA's emphasis to effectively use gravity and microgravity and other characteristics of the space environment to enhance an understanding of fundamental biological processes.

A major portion of the research activities in this project was directed towards gaining a better understanding of the vegetative propagation (root development) of sweetpotato stem cuttings under hypogravity (using clinorotation; actual space flight). Of particular interest was an examination of the amyloplast distribution in stems and roots to determine whether hypogravity alters distribution of starch granules, which may limit availability of carbohydrates to support root growth.

This report summarizes both ground-based (with clinostats) and space flight studies (Shuttle) at Tuskegee University on using sweetpotato stem cuttings to develop a database in preparation for future flight opportunities. This work was done under NASA Grant # NAG10-0209 through John F. Kennedy Space Center. Several preliminary experiments were conducted both in the greenhouse and growth chambers to ensure a consistent performance of the clinostats as well as to determine optimal growing conditions.

Project Objective: The objective of this project was to examine the vegetative propagation (root development) of sweetpotato stem cuttings under hypogravity (using clinorotation). This would be accomplished through an examination of the amyloplast distribution in stems and roots to determine whether clinorotation alters distribution of starch granules, which may limit availability of carbohydrates to support root growth.

Clinostat Studies: Sweetpotato stem cuttings of the breeding clone 'TU-82-155' were grown in autoclaved Phytagel (Sigma Inc.), a nutrient agrose medium impregnated with a modified half Hoagland nutrient solution and grown for 14 days. Two orientation treatments - (1) horizantal rotation (HR), (2) vertical rotation (VR) and two stationary controls, horizantal (HS) and vertical (VS) were used. Eight stem cuttings were inserted to a depth of approximately 2.5 cm into the Phytagel medium. Stem cuttings that were rotated vertically on the clinostats, and those that were not rotated had more fibrous roots that were thicker. These roots also showed a normal gravitropic response, i.e. they grew downward in response to gravity. In contrast, fibrous roots on sweetpotato stem cuttings that were rotated on a horizontal plane were shorter, thinner, and tended to curve upward parallel to the axis of rotation. Electron microscopy results indicate that amyloplasts (starch storage bodies in the cell) were evenly distributed in samples taken from stem cuttings that were rotated vertically and scattered among samples taken from stem cuttings that were rotated horizontally.

Space Flight Studies: Over the course of the grant we took advantage of two space flight opportunities with two commercial entities. In 1998 we leased three Liquid Mixing Apparatus (LMA) vials from Instrumentation Technology Associates, which had manifested commercial space on the STS-95 Space Shuttle flight. Unfortunately, the stem cuttings never grew, and actually died, so no useful data were obtained. In 1999 we flew sweetpotato stem cuttings as part of Bioserve Space Technologies Commercial Generic Bioprocessing Apparatus (CGBA) payload on STS-93. Twelve stem cuttings of TU-82-155 (6 cm long) containing 3 to 4 nodes were grown in each of two (one used as ground-based control) plant growth units in Phytagel impregnated with half Hoagland nutrient solution, and flown on Space shuttle Columbia for 5 days. Upon return to earth, fibrous roots were counted, length determined, and samples chemically fixed for electron microscopy evaluation. CO2, O2, and ethylene levels were determined post-flight. Both ground based and space flight cuttings generated fibrous roots. However, space flight cuttings generated a greater number and length of fibrous roots than ground-based controls. CO2 were marginally higher in the space flight containers, while ethylene levels were similar, and averaged < 10ppb. Amyloplast appeared sedimented in fibrous roots of ground-based and scattered in flight-grown plants. Results of carbohydrate analyses indicated substantial increases in carbohydrates in the stem cuttings grown in microgravity (glucose 171%, fructose 65%, sucrose 42%, and starch 214% greater) than the stem cuttings that were grown here on earth. This is finding is similar to those of others and could have been caused by microgravity-induced root zone hypoxia, as the air space within the container averaged 18.9% O_2. The O_2 availability to the roots within the growth media was not measured directly as this was technically difficult.

Publications/Presentations

Abstracts Published

Gamble, S.D., D.G. Mortley, C.E. Morris, C.S. Williams, J.W. Williams, C.F. Davis and P.A. Loretan. 1999. Effect of clinoration on sweetpotato stem cuttings. Gravitational and Space Biology Bull. 13:68.

Gamble, S.D., D.G. Mortley, C.E. Morris, C.S. Williams, J.W. Williams, and C.F. Davis. 1999. Clinorotation influences root growth and ultra structures of sweetpotato stem cuttings. Proc. National Alliance of NASAURC p.46.

Mortley, D.G., C.S. Williams, S.D. Gamble, C.F. Davis, and J.W. Williams. 1999.STS-93 space shuttle mission with sweetpotato stem cuttings. Gravitational and Space Biology Bull. 3:71

Mortley, D.G., C.S. Williams, C.F. Davis, and J.W. Williams. 2000. Effect of microgravity on root regeneration, ultrastructures, and carbohydrate content of sweetpotato stem cuttings. Gravitational and Space Biology Bull. 4:39.

Williams, C.S., D.G. Mortley, S.D. Gamble, C.F. Davis, S.D. Gamble and J.W. Williams, and P.A. Loretan. 1999. STS-95 space shuttle mission – a learning experience. Gravitational and Space Biology Bull. 3:71.

Williams, C.S., D.G. Mortley, .C.E. Morris, C.F. Davis, S.D. Gamble, and J.W. Williams, and P.A. Loretan. 2000. Microscopic analysis of swewetpotato roots propagated from stem cuttings maintained in either vertical or horizantal clinorotation. Gravitational and Space Biology Bull. 4:35.

Presentations

Gamble, S.D., D.G. Mortley, C.E. Morris, C.S. Williams, J.W. Williams, C.F. Davis. Effect of clinorotation on sweetpotato stem cuttings. 15th Annual Meeting American Society for Gravitational and Space Biology, Seattle, WA, November, 1999.

Gamble, S.D., D.G. Mortley, C.E. Morris, C.S. Williams, J.W. Williams, and C.F. Davis. Clinorotation influences root growth and ultra structures of sweetpotato stem cuttings. Proc. National Alliance of NASAURC Student Conference. Nashville , April 2000.

Mortley, D.G., C.S. Williams, S.D. Gamble, C.F. Davis, and J.W. Williams. STS-93 space shuttle mission with sweetpotato stem cuttings. 15th Annual Meeting American Society for Gravitational and Space Biology, Seattle, WA, November, 1999

Mortley, D.G., C.S. Williams, C.F. Davis, and J.W. Williams. 2000. Effect of microgravity on root regeneration, ultrastructures, and carbohydrate content of sweetpotato stem cuttings. 16th Annual Meeting, American Society for Gravitational and Space Biology, October 25-28, Montreal, Canada.

Williams, C.S., D.G. Mortley, S.D. Gamble, C.F. Davis, S.D. Gamble and J.W. Williams, and P.A. Loretan. 1999. STS-95 space shuttle mission – a learning experience. Gravitational and Space Biology Bull. 3:71.

Williams, C.S., D.G. Mortley, C.E. Morris, C.F. Davis, S.D. Gamble and J.W. Williams. Microscopic analysis of swewetpotato roots propagated from stem cuttings maintained in either vertical or horizantal clinorotation. 16th Annual Meeting, American Society for Gravitational and Space Biology, October 25-28, 200, Montreal, Canada.

Manuscripts in Preparation

Gamble, S.D. et al. Response of sweetpotato stem cuttings to clinorotation (MS Thesis; Life Support and Biosphere Science)

Mortley, D.G. et al. Influence of microgravity on root regeneration of sweetpotato stem cuttings and ultrastructure integrity. Life Support and Biosphere Science.